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ORIGINAL ARTICLE

Biosynthesis of nanosilver particles using extract of *Salmonella typhirium*

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Abstract Development of reliable and eco-friendly processes for the synthesis of metallic nanoparticles is an important step in the field of application of nanotechnology. One of the options to achieve this objective is to use natural factories such as biological systems. In the present investigation, we report the biosynthesis of silver nanoparticles employing extract of *Salmonella typhirium*. Silver nanoparticles were synthesized through the reduction of aqueous Ag^+ ion using the cell extract of bacterium *Salmonella typhirium* in the bright conditions. The synthetic process was fast and silver nanoparticles were formed within 30 min of silver ion coming in contact with the cell extract filtrate. UV–visible spectrum of the aqueous medium containing silver ion showed a peak at 415 nm corresponding to the plasmon absorbance of silver nanoparticles. The size of the nanoparticles was measured by dynamic light scattering (DLS) and the morphology of nanoparticles was observed using transmission electron microscopy (TEM).

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1. Introduction

Nanotechnology plays a very important role in many key technologies of the new millennium (Mandal et al., 2006). The application of nano-scale and nano-structure materials within range of 1–100 nm is an emerging area of nanoscience and nanotechnology. Nanomaterials may provide solutions to technological and environmental challenges in the areas of

solar energy conversion, catalysis, medicine, and water treatment (Sharma et al., 2009).

Nanoparticles of silver have many important applications that include: spectrally selective coating for solar energy absorption and intercalation material for electrical batteries, as optical receptors, polarizing filters, catalysts in chemical reaction, biolabeling and as antimicrobial agents (Ahmad et al., 2003).

For the production of nanoparticles, one needs to know the physical and chemical principles of nanoscale materials and also know how to commercialize them. Generally, metal nanoparticles can be prepared and stabilized by chemical, physical, and biological methods; the chemical approaches, such as chemical reduction, electrochemical techniques, photochemical reduction (Sharma et al., 2009; Jacob et al., 2007), pyrolysis (Qiaoxin et al., 2009) and physical methods, such as Arc-discharge and physical vapor condensation (pvc) (Tavakoli

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et al., 2007) are used. Living organisms have huge potential for the production of nanoparticles. Recently found that microorganisms have been explored as potential biofactory for the synthesis of metallic nanoparticles such as cadmium sulfide, gold and silver (Sharma et al., 2009; Vigneshwaran et al., 2007).

Researchers prefer biological synthesis, because distribution control of particles obtained from this method is better than other methods (Durán et al., 2007). In addition, this method involves no environment toxicity which is usually accompanied with other chemical methods (Kumar et al., 2007).

The first synthesis of silver nanoparticles by bacterium has been reported in 2000. Joerger et al. used *Pseudomonas stutzeri* bacterium (AG259) for silver nanoparticle synthesis with a size smaller than 200 nm (Joerger et al., 2000). In 2008, biosynthesis of silver nanocrystals by *Bacillus licheniformis* has been researched. Aqueous silver ions were reduced to silver nanoparticles when added to the biomass of *Bacillus licheniformis*. This was indicated by the change in color from whitish-yellow to brown. The probable mechanism for the formation of silver nanoparticles involves the enzyme nitrate reductase (Kalimuthu et al., 2008).

In 2009, it was investigated different visible-light irradiation's effects on the formation of silver nanoparticles from silver nitrate using the culture supernatant of *Klebsiella pneumoniae*. Also, the study experimentally investigated the liquid mixing process effect on silver nanoparticle synthesis by visiblelight irradiation (Mokhtari et al., 2009).

Finding rapid synthesis of silver nanoparticles by microorganisms is an important aspect. In this study, we used from extract of salmonella typhirium to reduce Ag^+ ions in an aqueous solution.

2. Materials and methods

2.1. Organism

Colloidal silver nanoparticle solution was prepared using the following previously described method. The bacterial culture,

Salmonella typhirium was obtained from Microbiology Laboratory, Tehran University, Tehran, Iran. Muller-Hinton broth (MHB) was prepared, sterilized, and inoculated with a fresh growth *Salmonella typhirium*. The cultured flasks were incubated at 37 °C for 24 h. After incubation, the cells were separated from the culture broth by centrifugation (5000 rpm) for 15 min and washed two times with deionized water to obtain some wet weight of cells (biomass).

The harvested cells were then resuspended in 50 mL of deionized water for 24 h. The cells were then removed by a 0.22-mm Durapore membrane filter (PVDF), and the filtrate obtained was extract of cell.

This solution prepared was a colorless liquid and was used for the reduction of silver sulfate. The filtrate was added separately to the reaction vessel containing silver sulfate at a concentration of 0.0005 M. The reaction between this filtrate and Ag^+ ions was carried out in bright conditions for 30 min. The products were characterized by Uv-vis spectroscopy, transmission electron microscopy (TEM), and dynamic light scattering (DLS) (see Fig. 1).

3. Results and discussion

For analytical study of the prepared sample, the amount of absorption within the wave length of 350–600 nm was observed by uv-vis spectroscopy. This technique has proven to be very useful for analyzing nanoparticles (Ahmad et al., 2003; Vigneshwaran et al., 2007). As illustrated in Fig. 2, a strong surface plasmon resonance was centered at ca. 415 nm. Observation of this strong but broad surface plasmon peak has been well documented for various metal nanoparticles, with sizes ranging widely from 2 to 100 nm (He et al., 2007; Ranganath et al., 2012).

In order to carefully study the size of the produced silver nanoparticles, dynamic light scattering analysis was used. As is seen in Fig. 3, average size of nanoparticle is about 129 nm and majority of nanoparticles has a size of 87 nm. Size distribution of silver nanoparticles is 30 nm which indicates narrow distribution of the produced nanoparticles. In addition,

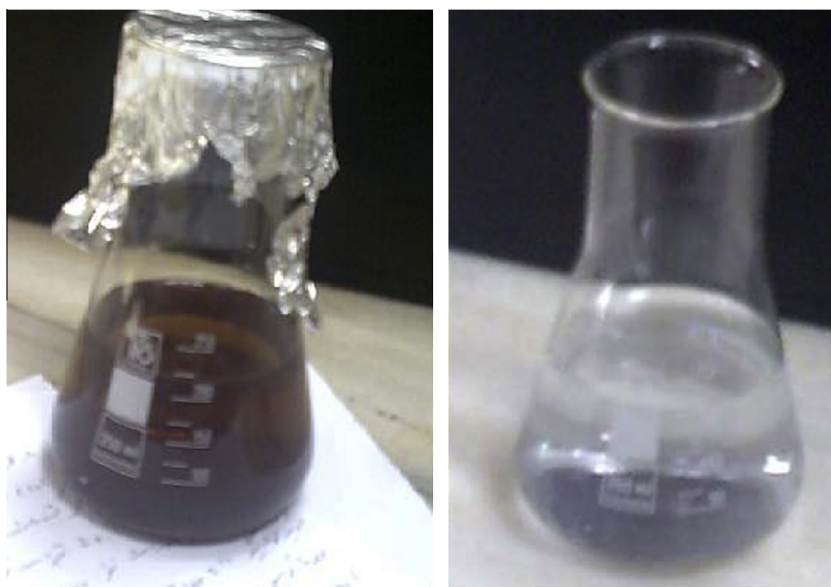


Figure 1 Solutions of silver sulfate (0.0005 M) before (right) and after (left) exposure to extract of *Salmonella*.

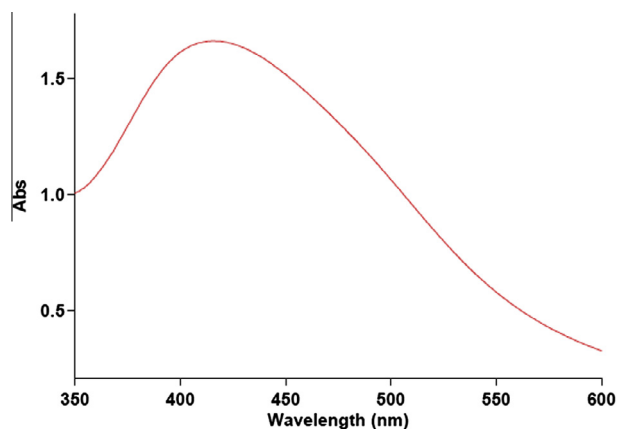


Figure 2 Uv-vis of Ag colloids. Spectra recorded after the addition of extract of cell (15 ml) to 100 ml of silver sulfate solution (0.0005 M).

tion, the largest nanoparticle volume (weight) was obtained at the size of 127 nm. In fact, silver nanoparticles with size of 127 nm had the largest amount of weight (Fig. 4).

At the end, silver nanoparticles synthesized by extract of *Salmonella* were studied by transitional electron microscopy (TEM) and images show and confirm silver nanoparticle production at nano-size. TEM images of silver nanoparticles are shown in Fig. 5.

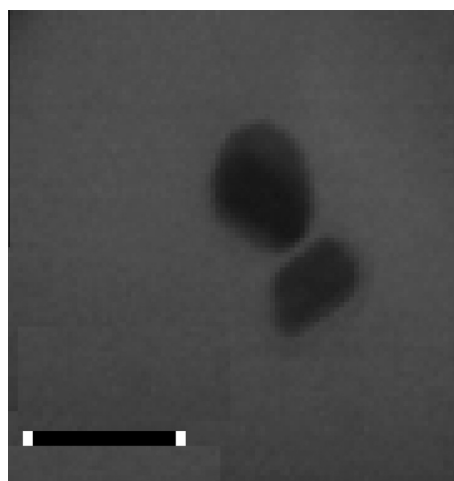


Figure 5 TEM of silver nanoparticles produced by extract of *Salmonella* (scale bare: 100 nm).

4. Conclusions

Silver nanoparticles in the range of 50–150 nm are synthesized by adding extract of *Salmonella typhirium* to silver sulfate. We showed that material released from interacell and cell wall of *Salmonella* lead to produce silver nanoparticles. The probable mechanism for the formation of silver nanoparticles involves

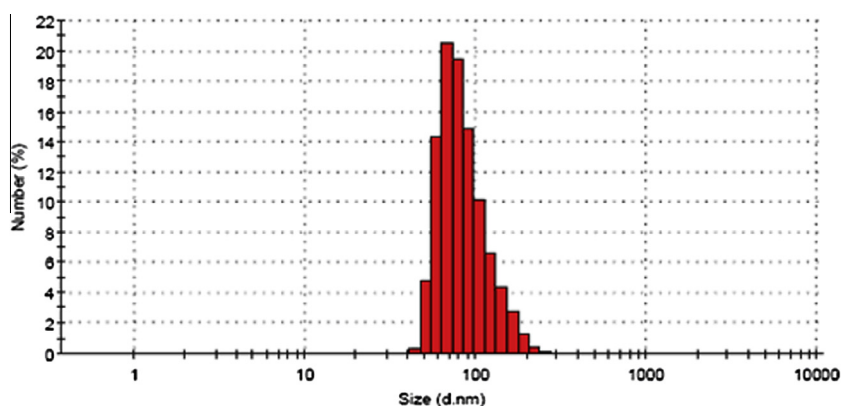


Figure 3 The curve of size distribution (silver nanoparticles) by number.

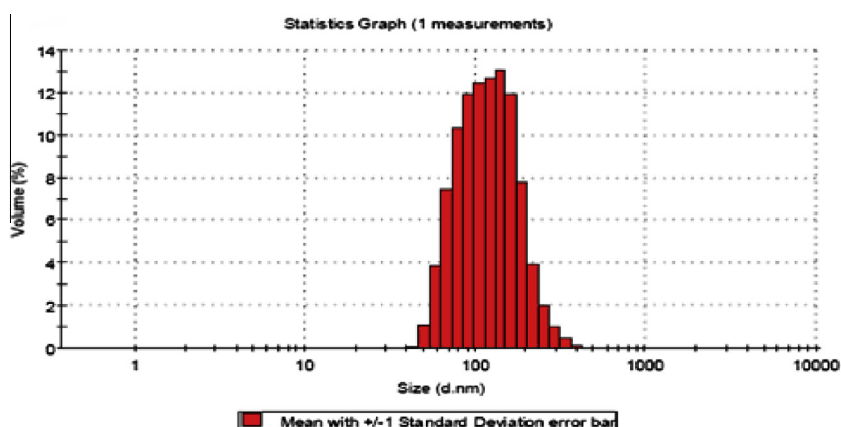


Figure 4 The curve of size distribution (silver nanoparticles) by volume.

the reductase enzymes and polysaccharides into cell and its wall. It seems the reductase enzymes released by *Salmonella typhirium* can reduce silver ions, although more research should take place in this area. This methodology could be used for synthesizing a number of metallic nanoparticles involving other metals with good size and shape morphology.

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